

Marking Scheme
BIOTECHNOLOGY (045)
Class-XII (2022-23)

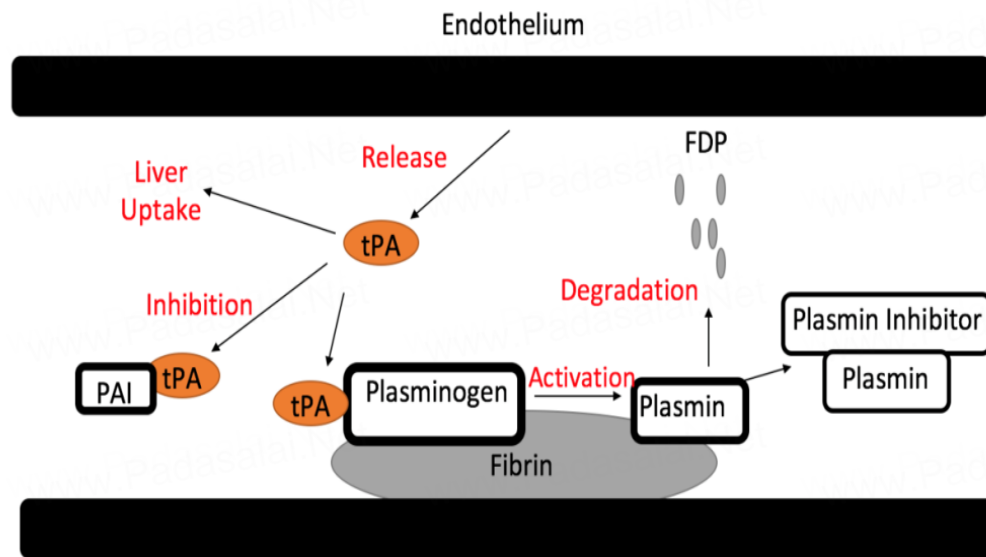
SECTION-A		
1	(a) Barnase protein	1
2	(c) Higher calcium and phosphorus content	1
3	(b) <i>Lithospermum erythrorhizon</i>	1
4	(a) In response to Internal and external changes the biochemical machinery of the cell could be changed.	1
5	(a) Encapsulating somatic embryos in calcium alginate beads	1
6	(c) Protein engineering	1
7	(d) Precision of delivery	1
8	(c) Explant culture	1
9	(d) Flexibility in choice of restriction enzyme	1
10	(c) Substitute another amino acid at position 222	1
11	(c) Slower, less safer and less specific	1
12	(c) BAC	1
13	A) Both Assertion and Reason are true and the reason is the correct explanation of the assertion	1
14	(C) Assertion is true but Reason is false	1
15	(A) Both Assertion and reason are true and reason is the correct answer for the assertion.	1

16	A) Both Assertion and Reason are true but the reason is not the correct explanation of the assertion	1
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SECTION-B

17	<p>Tissue Plasminogen Activator (tPA)</p> <p>Diagram /Flow chart (Either one)</p> <p>Production and mode of action of tPA.</p> <p>Production: A plasmid containing a t-PA gene insert is introduced into mammalian cells in a petri plate. These cells are then scaled up and transferred into a fermentor to produce tPA.</p> <p>Mode of Action: tPA is released from the endothelium. It activates Plasminogen to Plasmin. Plasmin then degrades Fibrin into FDP. tPA is also inhibited by PAI (Plasminogen Activator Inhibitor) and taken up by the liver.</p>	1
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Flow chart



18	<p>Somaclonal variations</p> <p>It helps in production of mutants e.g. disease resistance in Potato</p> <p style="text-align: center;">OR</p> <p>Organogenesis</p> <p>If auxins are high in the medium, it promotes rooting while if cytokinins are high, shoot formation is promoted.</p>	1+1
19	<p>G amino acid is most conserved</p> <p>A amino acid is most variable.</p>	1 1
20	<p>Essential amino acids and BCAA profile: Essential amino acids are those amino acids which have to be obtained from food and cannot be made in our cells.</p> <p>The branched chain amino acids (BCAA) are essential for the biosynthesis of muscle proteins. They help in increasing the bio-availability of high complex carbohydrates intake and are absorbed by muscle cells for anabolic muscle building activity.</p> <p>Biological value (BV) measures the amount of protein nitrogen that is retained by the body from a given amount of protein nitrogen that has been consumed. It has been observed that the BV of whey proteins is the highest compared to rice, wheat, soya and egg proteins.</p> <p>Protein efficiency ratio (PER)- PER is used as a measure of growth expressed in terms of weight gain of an adult by consuming 1g of food protein. The PER value of the following proteins are arranged in decreasing order- whey,milk, casein, soya, rice, wheat.</p> <p style="text-align: right;">(Any two)</p>	1 1
21	<p>a) Production of MoAb (0.5 mark)</p> <p>b) This technology has revolutionized the area of diagnostics and antibody-based therapies.</p> <p>1) The availability of monoclonal antibodies has helped in the early detection of many infectious diseases like hepatitis and AIDS.</p> <p>2) Therapeutic mAb –</p> <p>OKT3 Therapeutic mAb - Herceptin OKT-3 is monab-CD3, an immunosuppressant drug given intravenously to reverse the acute rejection of transplanted organs such as the heart, kidney and liver.</p> <p>Herceptin (trastuzumab) is a monoclonal antibody approved for therapy of early-stage breast cancer that is Human Epidermal growth factor Receptor 2-positive (HER2+). (1.5 marks)</p>	2

SECTION-C

22

(a) In chymotrypsinogen, the substrate binding site is blocked and hence the enzyme is inactive. In-situ activation of trypsin involves a proteolytic cut in chymotrypsinogen which results in a conformational change, exposing the substrate binding pocket.

(b) Asp 102, His 57 and Ser 195 lie in this order forming a charge relay;

The negatively charged aspartate carboxylate residue pulls the Ser –OH proton through His, leaving it with a negative charge Ser195 becomes acidic due to the unique constellation of the three amino acid residues because the protein has folded uniquely in space

1+2

OR

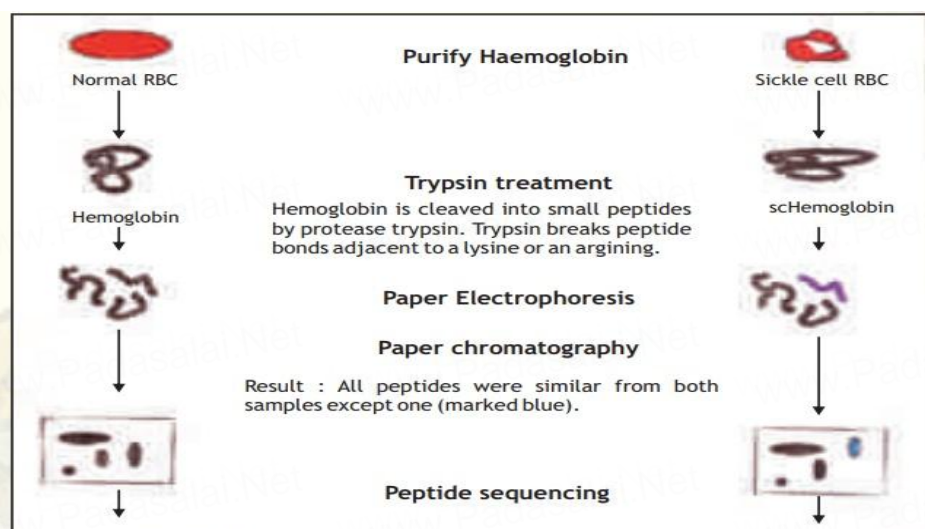


Fig. 6. Protein fingerprinting

 $\frac{1}{2} \times 6$ **Protein fingerprinting/ peptide mapping**

23

Crop	Gene	Improved Character
Canola	(A) Barnase Barstear	Hybrid production
Corn	(B) BtCryIA(c)	Insect Resistance
Cotton	(C) BtCryIA(c)	Insect Resistance
Papaya	(D) Coat protein	Virus Resistance
Potato	(E) BtCryIIIA & Coat protein	Insect & virus control
Soyabean	EPSP synthase	Weed control

 $\frac{1}{2} \times 6$

24	<p>Membrane integrity maintained</p> <p>Helps to maintain the shape and size of cells.</p> <p>Salt, glucose and amino acids (any two) are the major ingredients that determine osmolality of the medium.</p>	1x3
25	<p>(a) →BLAST search→ Find out→ homologous sequences in other organisms by looking for gene sequence of given proteolytic enzyme.</p> <p>(b) Look for conserved domain and find whether belongs to domain of Chymotrypsin or to other family of proteins</p> <p>(c) ALI database can be used for Phylogenetic (Evolutionary) analysis and alignment of proteins.</p>	1 1 1
26	<p>R.E. type II recognizes a specific DNA sequence and cut within the sequence generating sticky/flush ends. In recombinant DNA technology, we use type II RE as they are highly specific in their action.</p> <p>Alu I with the restriction site (One strand) 5' AGCT'3 and Sma I with the restriction site 5 'CCC GGG' 3 (flush ends) (One strand)</p> <p>The functions of a) Alkaline phosphatase b) DNA ligase.</p> <p>*The role of alkaline phosphatase is to prevent self re-ligation of the vector</p> <p>*The role of DNA ligase is to make 3'-5' phosphodiester bond.</p>	1 1 ½ ½
27	<p>: i) UniGene database</p> <p>ii) Homologene database</p> <p>iii. RefSeq database</p>	1 1 1
28	a) p BR 322	1
	<p>b) LEU2 gene codes for an enzyme required for the synthesis of amino acid leucine.</p> <p>Yeast cells having this plasmid can grow on a medium lacking leucine and hence can be selected e.g. Yep</p>	1 ½ ½

SECTION- D

29	<p>(a) The molecular ions are generated either by a loss or gain of a charge (e.g. electron ejection, protonation or deprotonation) 1</p> <p>(b) Mass spectrometry is used in- 1</p> <p>(i) Obtaining protein structural information such as peptide mass or amino acid sequence</p> <p>(ii) Identifying the type and location of amino acid modification within proteins. (any one)</p> <p>(c) (c)$m/z = (M + nH)^{n+} / n^+$ 2</p> <p>For $n=5$, $m/z = 10,000 + 5/5 = 2001$ For $n=4$, $m/z = 10,000 + 4/4 = 2501$ For $n=3$, $m/z = 10,000 + 3/3 = 3334.3$ For $n=2$, $m/z = 10,000 + 2/2 = 5001$</p> <p style="text-align: center;">OR</p> <p>(c) $m/z = (M + nH)^{n+} / n^+$ For $n=6$, $m/z = 20,000 + 6/6 = 3334.33$ For $n=7$, $m/z = 20,000 + 7/7 = 2858.14$</p>	
30	<p>a) As generation time is inversely related to specific growth rate, hence bacterial culture marked "X" with generation time 20s will proliferate rapidly. 1</p> <p>b) $n = 3.3 (\text{Log } 10^7 - \text{Log } 10^4)$ 1 $= 3.3 (7 - 4)$ $= 10$</p> <p>c) First calculate the number of divisions the population must have undergone to increase from 10^8 to 10^{14} in 4 hours. 2 $n = 3.3 (\text{Log } 10^{14} - \text{Log } 10^8)$ $= 3.3 (6)$ $= 19.8$ $t_d = 240 \text{ minutes} / 20$ $= 12 \text{ minutes}$</p> <p style="text-align: center;">OR</p> <p>c) (i) Measurement of Dry mass and Wet mass (ii) Using spectrophotometer (iii) Using Slide counting Chamber (iv) Using Coulter chamber (Any two)</p>	

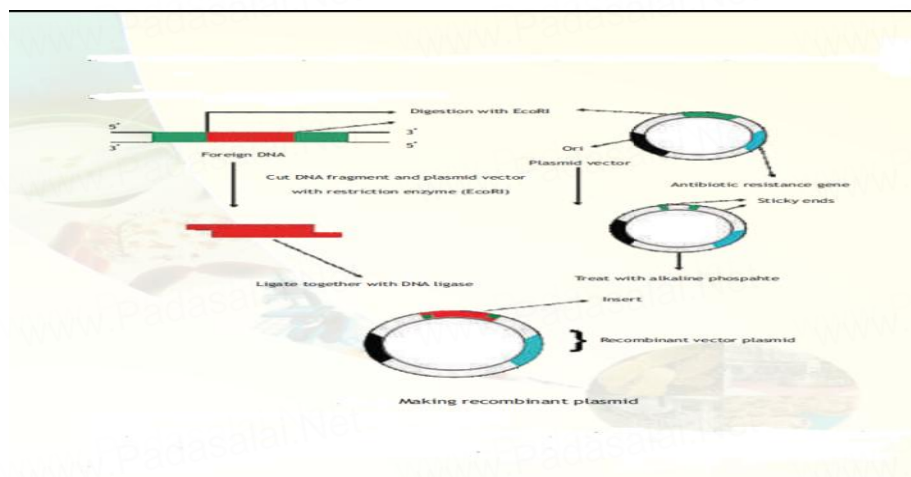
SECTION- E

31	<table border="1" data-bbox="248 283 1370 617"> <thead> <tr> <th>Proteins</th> <th>Animal cell line used</th> <th>Therapeutic use</th> </tr> </thead> <tbody> <tr> <td>Erythropoietin</td> <td>CHO cells</td> <td>Anemia</td> </tr> <tr> <td>Factor VIII</td> <td>CHO cells</td> <td>Hemophilia A</td> </tr> <tr> <td>Follicle Stimulating Hormone (FSH)</td> <td>CHO cells</td> <td>Infertility</td> </tr> <tr> <td>Interleukin 2 (IL 2)</td> <td>CHO cells</td> <td>Cancer therapy</td> </tr> <tr> <td>Monoclonal antibodies (mAbs)</td> <td>Hybridoma cells</td> <td>Cancer therapy & Autoimmune diseases</td> </tr> </tbody> </table> <p style="text-align: center;">OR</p> <p>(a) (i) A defined medium has known chemicals, of fixed composition and can support growth of selected cells. Serum is an essential component of animal cell culture media and is a source of growth factors and hormones. 2</p> <p>(ii) Anchorage dependent cells grow as adherent cells whereas anchorage-independent cells grow as suspension cultures.</p> <p>(b) Most common buffering system used to maintain pH in animal cell Culture is Bicarbonate-CO₂ system. Carbon dioxide from cells or atmosphere interacts with water and leads to drop in pH. 2</p> $\text{H}_2\text{O} + \text{CO}_2 \rightleftharpoons \text{H}_2\text{CO}_3 \rightleftharpoons (\text{H}^+) + \text{HCO}_3^{-1}$ <p>Increase in Bicarbonate concentration neutralizes the effect of increased Carbon dioxide according to the following equation:</p> $\text{NaHCO}_3 \rightleftharpoons \text{Na}^+ + \text{HCO}_3^{-1}$ <p>The increased HCO₃⁻ ions derive the above equation to its left until equilibrium is reached at pH 7.4 1</p> <p>Advantages :</p> <p>i) pH is important to maintain in balance/ enzyme functions/ binding of hormones/growth factors to cell surface receptors/Ion balance (Any two)</p>	Proteins	Animal cell line used	Therapeutic use	Erythropoietin	CHO cells	Anemia	Factor VIII	CHO cells	Hemophilia A	Follicle Stimulating Hormone (FSH)	CHO cells	Infertility	Interleukin 2 (IL 2)	CHO cells	Cancer therapy	Monoclonal antibodies (mAbs)	Hybridoma cells	Cancer therapy & Autoimmune diseases	½ x10
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32	<p>(a) TaqDI 1</p> <p>(b) 5' AATGC 3' and 5' GATTC 3' 1</p> <p>(c) Palindromic means the DNA sequence reads same when read from 5' to 3'. The Restriction enzyme is a homodimer. ½</p> <p>As it cuts both the strands of DNA simultaneously in 5' to 3' direction. ½</p> <p>(d) Foreign DNA can be inserted into bacteriophage single stranded, circular DNA of 6407 bp without disrupting any of the essential genes 2</p> <p>M13 is a filamentous phage which infects E. coli having a pilus (protrusion) which is selectively present in cells containing a F plasmid (called F+ cells).</p>	1 1 ½ ½ 2																		

OR

2

a)



b) Replica plating.

 $\frac{1}{2}$

- Host cells are first plated (master plate) on solid media with the desired antibiotic overnight.
- Velvet paper is aligned, pressed on master plate.
- With the same alignment it is pressed onto the replica plate.
- Keep it overnight, transformed colonies will not grow in replica plate
- The colonies having insert can easily be scored off from master plate by comparing the two plates.

 $\frac{1}{2} * 5$

33

- (a) Recombinant insulin is an intracellular protein so we need to process the cell mass and not the fermentation broth.
- (b) Strain improvement is done in order to maximize metabolite production by:
- Mutant selection : There are two methods - Physical method & Chemical Method
 - Genetic engineering
- (c) i) It has strong inducible promoters
- It is capable of making post-translational modifications similar to those performed by human cells
 - Downstream processing is simpler as Pichia does not secrete its own proteins into the fermentation medium.
- (Any two)

1

1

1

1x2

OR

	<p>a) <u>Use of shake culture and Use of baffle flask</u></p> <p>Baffle flask: One of the simplest ways is to produce a V- shaped notch or indentation in the sides of the flask. Such flasks are called baffle flasks . This improves the growth of the microbes by improving the efficiency of oxygen transfer due to increased turbulence of the agitated culture medium.</p> <p>Shakers: Continuous agitation of the culture medium also greatly improves the efficiency of the oxygen transfer and this improves the growth of the microbes. In the laboratory, this is done by the use of shakers . Shakers may be end-to-end type or rotatory type. These may be designed for use at the ambient temperature or in a controlled temperature environment (incubator shaker).</p>	1x2
	<p>b)</p> <ol style="list-style-type: none"> 1. Production of whole microbial cells (for food, vaccines) 2. Production of primary metabolites (acids, alcohol) 3. Production of secondary metabolites (antibiotics) 4. Biotransformation reactions (enzymatic, steroid) 5. Exploitation of metabolism (microbial leaching, biodegradable waste treatment) 6. Synthesis of recombinant proteins (therapeutic proteins) Bioremediation/fermented food items/ recombinant proteins (Any two) 	1x2
	<p>c) Viable Plate Count is the best method since it does not count dead microbial cells.</p>	1